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Loss of Protein, Immunoglobulins, and Electrolytes in Exudates From Negative Pressure Wound Therapy

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Background: A relatively new technology in wound care, negative pressure wound therapy (NPWT), has become widely used for the management of open abdomens and soft tissue wounds and provides a means to collect wound exudate to quantify protein loss. *Methods:* A prospective observational study was conducted in surgical, trauma, or burn patients (8 patients with open abdomens and 9 patients with acute soft tissue wounds on NPWT). NPWT exudate was collected and assayed to characterize loss of protein, electrolyte, and immunoglobulins over multiple days of NPWT. *Results:* Total protein was present in open abdomen NPWT exudate, 2.9 ± 0.9 g/dL. In the soft tissue wound exudate, a similar mean concentration was found, 2.59 ± 0.6 g/dL (P = .34). Exudate concentrations of albumin, urea nitrogen, immunoglobulins, and electrolytes between wound types were also not significantly different. There were significant (P = .03) differences in the median volume of exudate, 1031 mL/d for open abdomens in contrast to 245 mL/d soft tissue wounds. Therefore, 24-hour losses of proteins and electrolytes were greater in patients with open abdomens than soft tissue wounds. Mean total protein loss was 25 ± 17 g/d for open abdomens and 8 ± 5 g/d for soft tissue wounds. *Conclusion:* There are significant losses of proteins in wound exudate. As there is no significant difference in the concentration of total protein between wound type, the rate of loss may be calculated as 2.9 g/dL times the volume of wound exudate. The rate of protein loss from wounds is similar to the presently assumed insensible loss rate of 12-25 g/d. (*Nutr Clin Pract*. 2010;25:510-516)

Keywords: negative-pressure wound therapy; wounds and injuries; exudates and transudates; carrier proteins, nutrition requirements, nitrogen

The hypermetabolic and catabolic state of critically ill surgical, trauma, and burn patients and subsequent importance of nutrition support has been well described.[1-6] Nutrition support, with emphasis on appropriate caloric and protein intake, is an important component in the overall management of the surgical and trauma patient.[1-6] Often patients require increased protein and calories as well as additional nutrition support to promote adequate wound healing.[7] Failure to consider protein loss from wounds leads to inadequate nutrition support due to underestimation of protein requirement, causing significant cumulative protein deficits and, ultimately, a poor clinical outcome associated with protein malnutrition. At present, the protein loss from a wound is at best an estimate, and calculation of replacement is not validated. Nitrogen (N) balance equations are commonly used to assess protein requirements, but the standard N balance equation does not account for abnormal N loss through wounds or burns.[6] As 6.25 g of protein = 1 g of N, a factor of up to 4 g N (25 g of protein) is used to account for desquamated skin, stool, or non-urea N loss.[6]

A relatively new technology in wound care, negative pressure wound therapy (NPWT), has become widely used for the management of open abdomens and soft tissue wounds and provides a means to collect wound exudate to quantify protein loss.[8-17] Determining the amount of protein lost from the exudate produced from open abdomens or soft tissue wounds using NPWT may improve the assessment of protein requirements, optimizing nutrition management, and promoting wound healing.

We hypothesize that patients with open abdomens or soft tissue wounds managed by NPWT have significant amounts of protein loss from wound exudate. We aimed

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to compare protein loss from 2 extreme situations (open abdomen vs soft tissue wound) in which NPWT would be used. In addition to determining protein loss, we also aimed to characterize the electrolyte and immunoglobulin composition of the NPWT exudate.

Methods

Design

All procedures were reviewed and approved by the Brooke Army Medical Center Institutional Review Board. We conducted a prospective, observational study to quantify the amount of protein present in the exudate collected from NPWT. The study was conducted in the surgical, trauma, and burn intensive care units (ICUs) of a level I trauma center. Between May 2008 and January 2009, surgical and trauma patients admitted to the ICU were screened for potential study enrollment. Consecutive patients on NPWT expected for >3 days with open abdomens or acute soft tissue wounds were enrolled. Patients <18 years of age, pregnant, or treated with silver-containing dressings were excluded.

Procedure

Once a patient met inclusion criteria, samples of exudate (≤20 mL) were collected during the designated 24-hour period. Sampling frequency was in relation to the day NPWT was initiated, which was designated as day 1. Wound exudate was collected daily on days 1, 2, and 3, and then every other day until day 9 or until NPWT was discontinued. Convenience based on clinical care was a consideration in the process; therefore, exudate was not collected if the patient was unavailable (eg, when undergoing operative procedures). On these occasions, it was not always possible to obtain adequate specimen amount on the designated day.

The wound exudate collection system used was an integrated specimen trap, similar to that developed by Banwell and Teot.[8] A sputum trap was inserted in the line draining the wound fluid into the NPWT wound exudate canister.

Exudate samples were centrifuged at 2000 × *g* for 10 minutes to separate any blood cells, fibrin, or cellular wound debris that may have been present. Samples were frozen at –70°C until the assays were performed. After thawing, samples were gently mixed before analysis. Exudate samples were assayed for total protein, albumin, urea N concentration, sodium, potassium, chloride, phosphorus, magnesium, calcium, and immunoglobulins (IgA, IgG, and IgM). Chemistry and immunoglobulin analysis was performed using the Dimension™ Xpand™ Clinical Chemistry System, Siemens Healthcare Diagnostics (Deerfield, IL). Samples with total protein concentrations <2.0 g/dL were assayed in duplicate using the Bio-Rad Protein Assay, microassay procedure (Bio-Rad Laboratories, Richmond, CA).

Patient demographics, diagnosis, mechanism of injury, and serum concentrations of total protein, albumin, and blood urea nitrogen (BUN) were recorded for each patient enrolled. Also recorded were daily exudate volume, wound size (estimated or measured per planimetry). The planimetry method involved using a sterile sheet of plastic measured for surface area and then weighed. The wound or open abdomen was traced on the plastic sheet and cut accordingly. The "wound size" cut piece of plastic was weighed and compared with the precut plastic sheet. The percentage weight difference was used to assess surface area of the wound, as surface area and weight of the precut plastic sheet were previously determined. Daily loss for each analyte was calculated by multiplying the concentration by corresponding daily exudate volume. Nutrient intake was recorded and 24 hour urine urea nitrogen (UUN) was measured. When applicable, corticosteroid administration, insulin regimen, intravenous albumin administration, and continuous renal replacement therapy (CRRT) were recorded. None of the patients enrolled received anabolic steroids, immunoglobulin G (IgG), or growth hormone.

Statistical Analysis

Descriptive statistical analysis for the data includes mean and standard deviations and, when appropriate, median and interquartile ranges for nonparametric data. The open abdomen and soft tissue wound groups were compared using a Student's t test for the normal data and Wilcoxon 2-sample test for nonparametric variables. Correlations used a Spearman test. All tests for significance were 2-tailed, with $\alpha = .05$.

Results

Exudate samples (n = 26) were collected from 8 open abdomens, and 29 exudate samples were collected from 12 soft tissue wounds. We were unable to collect the planned 6 samples per patient for all patients enrolled because of issues such as operative procedures or low wound exudate volumes. Demographic and diagnosis data for each group are presented in Tables 1 and 2. There were no differences in age between groups. For soft tissue injuries, more than 1 wound was studied in 3 patients.

Patient Treatment Data

In the open abdomen group, 2 patients received corticosteroids, 5 patients were administered intravenous

Table 1. Patient Demographics

	Open Abdomens	Soft Tissue Wounds
No. of patients	8	9
Age, y	48.6 ± 18.3	42.4 ± 21
Weight, kg	84.3 ± 24.3	84.4 ± 16
Gender, n, F/M	2/6	0/9
No. of wounds	8	12
No. of exudate samples	26	29

Table 2. Cause of Injury or Surgery and the Number of Patients for Each

Open Abdomens	Soft Tissue Wounds (11 extremity, 1 flank)
Pancreatic cancer, 1	Explosive device injury, 4
Blunt trauma, pelvic fracture, 1	Penetrating trauma, gunshot wound, 5
Motor vehicle collision, liver laceration, 1	Motorcycle collision, traumatic right above-the-knee amputation, 1
Bowel surgery for obstruction, 2	Pedestrian vs motor vehicle collision, traumatic left above-the-knee amputation, traumatic right foot wound, 1
Penetrating trauma, gunshot wound, 2	
20-foot fall, 1	

albumin, 1 patient was placed on CRRT, and 7 patients were treated with insulin (either sliding scale or intensive insulin therapy). All of the open abdomens studied were surgical or trauma patients. Kinetic Concepts Inc (San Antonio, TX) KCI NPWT was used in 7 patients; wall suction was used for 1 patient. One patient's NPWT pressure was 50 mm Hg, another patient's NPWT pressure was 184 mm Hg, and the remaining patients' NPWT pressures were 125 mm Hg. For nutrition intake, 3 of these patients received parenteral nutrition (PN), 1 patient received enteral feeds, and the remaining 4 patients received nothing by mouth; however, calories provided from propofol were included. Average daily intake was 1329 ± 1123 kcal and 51 ± 54 g protein. Of the 4 "fed" patients, the average intake was 1885 ± 1019 kcal and 80 ± 46 g protein. One of these patients received glutamine. Patients who received nothing by mouth had variable amounts of exudate output—as low as 175 mL in 24 hours but as high as 2250 mL in 24 hours. Therefore, receiving nothing by mouth did not appear to affect exudate volume output. There did not appear to be any significant differences in the concentration of proteins, electrolytes, or immunoglobulins in the exudates of the fed patients vs those receiving nothing by mouth, although our numbers were too small to draw any statistical conclusions.

In the soft tissue wound group, no patient received corticosteroids or was placed on CRRT; 2 patients were administered intravenous albumin, and 5 patients were treated with sliding scale insulin. KCI NPWT was used in 8 patients; wall suction was used for 1 patient. In this group, one patient's NPWT pressure was 150 mm Hg, the remaining soft tissue wounds on NPWT pressures were 125 mm Hg. Average daily nutrition intake (this includes only the patients receiving enteral nutrition or PN) was 911 ± 825 kcal (11 kcal/kg average actual body weight of 84 kg) and 53 ± 49 g protein. Three patients were receiving enteral feeds (one with PN), 3 were receiving regular diets, and 3 received nothing by mouth during the study period. Of the 3 patients who received nothing by mouth, the study period did not exceed 2 days. Enteral feeding or PN data were used in nutrition intake as calorie count data were not available for the patients receiving nothing by mouth. Two of these patients received glutamine.

We recorded CRRT, intravenous albumin, corticosteroids, and insulin use in our patients because all of these treatments can affect fluid flux between body compartments. Additionally, CRRT affects BUN levels and intravenous albumin affects serum albumin levels. The small number of patients receiving these therapies did not allow us to reach any statistical conclusions regarding how these therapies might affect wound exudate output or protein loss in the exudates.

Wound Exudate

Mean daily abdominal fluid output from the open abdomen group was 900 ± 547 mL/d (range, 120-2250 mL). Soft tissue wound exudate daily output volume was 359 ± 246 mL/d (range, 14-1100 mL). Median 24-hour exudate output for the soft tissue wounds was 245 mL/d, significantly less ($P = .03$) than observed for open abdomen loss, median 1031 mL/d. The difference between median wound surface area (543 cm^2 open abdomens vs 290 cm^2 soft tissue wounds) was not statistically significant ($P = .15$). The difference between median exudate volume in relation to wound surface area was not statistically significant ($P = .43$): 0.91 mL/cm^2/d in soft tissue wounds vs 1.23 mL/cm^2/d in the open abdomen group. Median total protein loss per wound surface area was 39 mg/cm^2/d in the open abdomens and 20 mg/cm^2/d in the soft tissue wounds ($P = .45$).

There were no significant differences between open abdomen and soft tissue wounds in the concentrations of total protein, albumin, or urea nitrogen in wound exudate (Table 3). Because of the greater volume of exudate from open abdomens, the loss of these analytes was significantly increased, by >3-fold (Table 3).

Table 3. Total Protein, Albumin, and Urea Nitrogen Concentrations and 24-Hour Loss in Negative Pressure Wound Therapy Exudates

	Open Abdomens	Soft Tissue Wounds	*P* Value
Albumin, g/dL	1.3 ± 0.4	1.0 ± 0.4	.079
Total protein, g/dL	2.9 ± 0.9	2.59 ± 0.6	.347
Urea nitrogen, mg/dL	25.0 ± 14.0	18.3 ± 6.6	.233
24-h loss of albumin, g	11.0 ± 7.0	3.2 ± 2.1	.017
24-h loss of total protein, g	25.0 ± 17.0	8.4 ± 5.2	.029
24-h loss of urea nitrogen, mg	221.0 ± 167.0	60.0 ± 40.4	.029

Values are mean ± standard deviation.

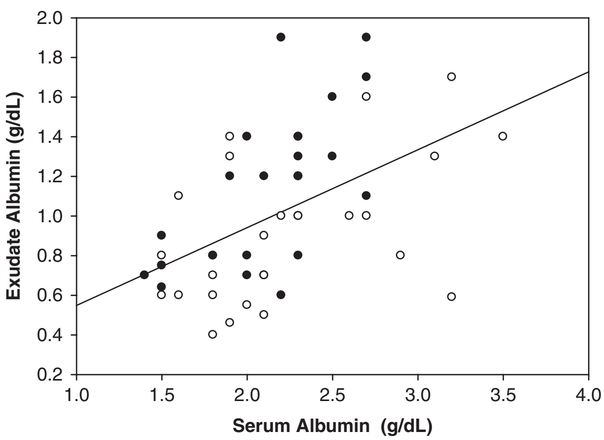

Figure 1. Exudate albumin concentration vs serum albumin $r = 0.52$, $P < .0001$, n = 51. Solid dots, open abdomens. Open dots, soft tissue wound.

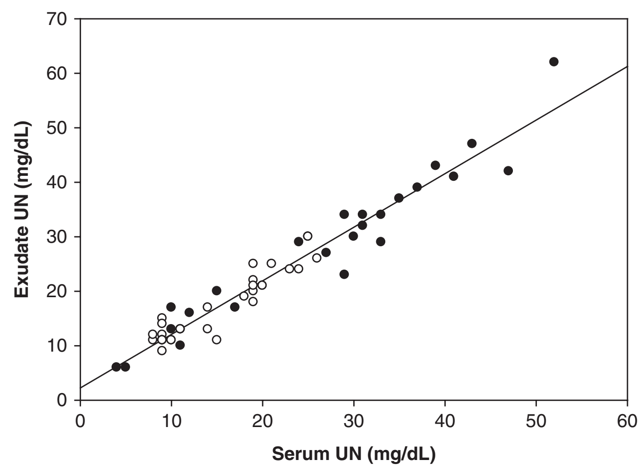

Figure 2. Exudate urea nitrogen vs serum urea nitrogen. $r = 0.95$, $P < .0001$, n = 55. Solid dots, open abdomens. Open dots, soft tissue wounds.

In the open abdomen patients, the mean serum total protein (TP) was 4.5 ± 0.8 g/dL, albumin was 2.3 ± 0.3 g/dL, and BUN was 22 ± 14.7 mg/dL. In the soft tissue wound group, serum TP was 4.8 ± 0.6 g/dL, albumin was 2.3 ± 0.6 g/dL, and BUN was 16 ± 6.1 mg/dL. Serum values were not significantly different between groups. There were positive correlations between serum and exudate for albumin ($r = 0.52$, $P < .0001$, n = 51); however, with an r^2 of 27%, this is a weak association (Figure 1). The positive correlation between serum and exudate for UN ($r = 0.95$, $P < .0001$, n = 55) is shown in Figure 2. No correlation was found for TP ($r = 0.12$, $P = .40$, n = 47). Mean TP (2.7 ± 0.98 vs 4.5 ± 0.73 g/dL) and albumin (1.0 ± 0.39 vs 2.2 ± 0.48 g/dL) levels were significantly lower in exudate compared with serum, whereas mean urea nitrogen (22 ± 11.7 vs 20 ± 11.5 mg/dL) was slightly higher.

Analysis of variance for the immunoglobulin data demonstrates significant difference only for 24-hour loss of IgA and IgM, with the open abdomen subjects having higher loss for both types of immunoglobulins (Table 4). In a similar pattern to the other analytes studied, concentrations for the electrolyte concentrations were not significantly different between the open abdomen and soft tissue wounds (Table 5). However, the daily loss of each electrolyte was significantly increased in exudate from open abdomens.

Discussion

Wound protein loss appears dependent upon the volume of exudates, as there were no concentration differences in proteins between open abdomens and soft tissue wounds. Although the difference in volume of exudate per wound surface area between groups may not have been statistically significant, exudate volume appears to be dependent on the wound area as the open abdomens had a greater rate of exudate formation for a given wound surface area. The use of the volume exudate collected in the course of NPWT as a means of estimating protein/nitrogen loss appears warranted.

Table 4. Immunoglobulin Concentration and 24-Hour Loss in Negative Pressure Wound Therapy Exudate

	Open Abdomens				Soft Tissue Wounds				
	Mean ± SD	25th Percentile	Median	75th Percentile	Mean ± SD	25th Percentile	Median	75th Percentile	*P* Value
IgA, mg/dL		56.6	79.0	89.6		33.0	58.5	143.8	.054
IgG, mg/dL		209.8	264.3	366.3		203.7	316.8	560.8	.073
IgM, mg/dL		18.1	28.3	64.7		13.0	15.6	21.3	.074
IgA, 24-h loss, mg	640.8 ± 445.7				249.9 ± 185.7				.044
IgG, 24-h loss, mg	2506.5 ± 1833.3				1036.1 ± 570.8				.059
IgM, 24-h loss, mg		128.5	322.8	444.4		27.1	45.0	91.4	.009

IgA, immunoglobulin A; IgG, immunoglobulin G; IgM, immunoglobulin M; SD, standard deviation. Concentration results of all the immunoglobulins were not normally distributed data (as was the 24-h IgM loss); therefore, median is used for analysis. The data for 24-h loss for IgA and IgG were normally distributed; therefore, mean is used for analysis.

Table 5. Electrolyte Concentrations and 24-Hour Loss in Negative Pressure Wound Therapy Exudate

	Open Abdomens	Wounds	*P* Value
Sodium, mEq/L, mean concentration	141.1 ± 7.6	142.3 ± 9.8	.83
Sodium, median 4-h loss, mEq (25th, 75th percentiles)	1492.1 (520, 1786)	361.2 (260, 767)	.034
Chloride, mEq/L, mean concentration	108.0 ± 6.0	107.8 ± 8.7	.95
Chloride, median 24-h loss, mEq (25th, 75th percentiles)	1133.0 (396, 1375)	276.0 (197, 572)	.034
Potassium, mEq/L, mean concentration	4.3 ± 0.57	4.5 ± 0.5	.52
Potassium, mean 24-h loss, mEq	37.1 ± 22.2	15.1 ± 8.5	.027
Phosphorus, mg/dL, mean concentration	3.3 ± 0.7	3.3 ± 0.9	.88
Phosphorus, mean 24-h loss, mg	26.7 ± 13.4	10.6 ± 6.1	.01
Magnesium, mg/dL, mean concentration	1.9 ± 0.3	1.9 ± 0.2	0.25
Magnesium, mean 24-h loss, mg	17.2 ± 10.2	6.3 ± 3.8	.02
Calcium, mg/dL, mean concentration	6.6 ± 0.9	6.1 ± 0.7	.20
Calcium, median 24-h loss, mg (25th, 75th percentiles)	72.0 (24, 90)	15.2 (12, 27)	.025

Of all the electrolytes in Table 5, only the 24-hour sodium, chloride, and calcium loss data were not normally distributed; therefore, median was used to analyze these data.

In 2007, Cheatham et al[18] reported on protein loss in open abdomens treated with the "vacuum pack" technique. They describe the vacuum pack technique as a dressing that incorporates the use of multiple-hole suction catheters placed on each side of the open abdomen under an occlusive dressing. This allows collection and quantification of all abdominal fluid removed from the abdominal cavity. From their data, they recommended that a nitrogen value of 2 g/L of abdominal fluid loss be considered for nitrogen balance equations.[6] Our study of open abdomen wounds suggests that this recommendation may not be adequate when NPWT is used. Our nitrogen concentration, when estimated from protein concentration (protein concentration/6.25), results in an N concentration of 4.6 g/L, a value twice their recommendation. Of interest, Cheatham et al found the daily rate of nitrogen loss from open abdomen exudate to be 3.5 g/d. In the present study, mean protein loss from open abdomens was 25 g/d or 4 g/d of nitrogen. The similar daily loss suggests differences in the volumes of exudate resulting in the concentration differences (higher exudates volumes were noted from the results reported by Cheatham et al compared with our results). We speculate that the difference between therapies, NPWT vs "vacuum pack" technique, could account for the difference in protein concentrations.

Waxman et al[19] described a significant protein loss in burn wounds. These investigators rinsed burn dressings and measured total protein and albumin in the water wash. They reported an average protein loss the first postburn week of 12 mg/cm^2/d. Our data showed amounts of protein loss greater than theirs, 46 ± 57 mg/cm^2/d in our soft tissue wounds. The difference may be due to the application of NPWT with our patients, leading to more protein being drawn from the wound, or the inclusion of patients with other types of soft tissue injury other than

burns. Also, it is possible that when burn dressings are rinsed, the sample may not include all of the protein that is lost. Furthermore, with NPWT, the volume of exudate is readily available, allowing rapid estimation of loss as protein concentration appears to be consistent irrespective of wound type in the 2 groups we studied.

Serum immunoglobulin levels were not obtained in the present study. Expected serum concentrations are: IgA 50-350 mg/dL, IgG-500-1500 mg/dL, and IgM 50-400 mg/dL.[20] Median concentrations of immunoglobulins in wound exudate fell in or below the lower range of normal plasma levels. The lower concentrations of immunoglobulins in wound exudate appear to mimic the relationship of other proteins in exudate to serum concentrations. Lehnhardt et al[21] reported differences among immunoglobulins as to their relationship to serum concentrations. Lehnhardt et al commented that immunoglobulins are the most important part of the humoral immunologic system and speculated that immunoglobulin supplementation, in appropriate amounts, might reduce infection rates. Being that little is known regarding immunoglobulin loss in NPWT, we chose to investigate this in the present study. We found that IgG in wound exudate was increased and IgM was decreased. In the present study, there appears to be a consistent pattern: immunoglobulin concentrations in wound exudate were below normal serum levels.

Unlike the proteins, electrolyte concentrations of exudates samples were similar to serum levels commonly reported in critically ill surgical, trauma, or burn patients. In the exudates of burn patients, Berger et al[22] found that magnesium loss was 16 mEq/d and phosphorus loss was 11 mEq/d. These values are similar to those observed in soft tissue wounds in the present study (Table 5). We noted that open abdomen wounds had higher daily losses of potassium, phosphorus, magnesium, and calcium than the soft tissue wounds, due to higher volumes of average exudate loss from the open abdomen. When combined with expected GI and urinary electrolyte loss, the additional loss of electrolytes through wound exudate is a contributor to electrolyte depletion and derangements and should be considered in estimations of electrolyte replacement.

This study has some limitations. NPWT is used on a variety of different wounds, and we looked at only 2 types: acute soft tissue wounds and open abdomens. Although our sample size was small, we were able to identify differences in protein loss with increasing exudate volumes. We would have preferred more exudate samples per patient also, but frequent unavailability of the patient (due to operative procedures, etc) limited our ability to accomplish this.

We noted significant associations of wound exudate albumin and urea nitrogen concentrations with serum levels (Figures 1 and 2). For albumin and total protein, the exudate concentrations were reduced, suggesting a greater flux of fluids, but not large proteins. Albumin accounts for about 55% of serum total protein. This is supported by the concentrations of electrolytes and urea nitrogen of exudate being similar to those of serum. The absence of an association of serum total protein and wound exudate suggests that other sources contribute to protein levels in the exudate. These sources could be damaged cellular components at the wound site as well as secondary inflammatory processes. This emphasizes the need to account for total protein loss from wound exudate.

Conclusions

Exudates from an open abdomen or soft tissue wounds on NPWT comprise a significant loss of protein, which should be considered when assessing protein requirements. Similar losses were noted for immunoglobulins and electrolytes. Losses were predominantly the result of differences in exudate volume rather than changes in concentrations. If unable to measure protein from wound exudate, clinicians should estimate protein or N loss when assessing N balance. In our patient population, irrespective of the wound type, protein/nitrogen loss can be readily estimated based upon the exudate volume and an assumed protein or nitrogen concentration of 2.9 g/dL or 4.6 g/L, respectively.

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Erratum

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The author order should have been:
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